Time course of structural adaptations in chronic AV block dogs: evidence for differential ventricular remodeling

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Verduyn, S. Cora, Christian Ramakers, Gabriel Snoep, Jet D. M. Leunissen, Hein J. J. Wellens, and Marc A. Vos. Time course of structural adaptations in chronic AV block dogs: evidence for differential ventricular remodeling. Am J Physiol Heart Circ Physiol 280: H2882–H2890, 2001.—To determine the nature and time course of biventricular hypertrophy and concomitant electrical and mechanical changes after creation of complete atrioventricular block (CAVB), six adult dogs (22–30 kg) were subjected to serial magnetic resonance imaging (MRI) and electrophysiography. After 6 days of CAVB, left ventricular (LV) mass, ejection fraction (EF), and Q-T time at a paced rhythm of 60 beats/min were already significantly increased. Maximal values were reached within 14–21 days of CAVB: LV mass, from 116 ± 11 to 143 ± 12 g; right ventricular (RV) mass, from 40 ± 3 to 55 ± 6 g; EF, from 68 ± 6% to 86 ± 5%; and Q-T time, from 285 ± 25 to 330 ± 35 ms, all P < 0.05. Cardiac output returned to baseline at day 14. End-diastolic wall thickness increased only in the RV, in which angiotensin type 1 (AT1) receptor mRNA expression was significantly greater. The autopsy correlated well with the MRI results (r = 0.98, P < 0.01). In conclusion, electrophysiological, mechanical, and structural adaptation processes after bradycardia-induced volume overload develop rapidly and are completed within 3 wk. The degree of hypertrophy was greater in the RV, which was associated with an increase in AT1 receptor mRNA.

biventricular hypertrophy, 2) increased inotropic performance at the slow idioventricular heart rate, and 3) nonhomogenous lengthening of the ventricular action potential duration (APD). There are, however, important differences in this adaptation process when the ventricles are compared, e.g., the right ventricular (RV) weight increase is more obvious than that in the left ventricular (LV), whereas the increase in LV APD is more than that in RV APD, leading to a more pronounced dispersion of repolarization. All these findings, which were confirmed at the cellular level (32), contribute to the observed enhanced susceptibility for afterdepolarizations and triggered arrhythmias in these dogs (34).

Structural remodeling in this animal model has been reported earlier because dogs with chronic CAVB have been tested experimentally since the beginning of the 20th century (3, 7). However, the time course and background of this hypertrophy process are largely unknown. In addition, we investigated whether the nature and degree of the hypertrophy are similar for the RV and LV and whether the time frame of the structural changes parallels mechanical and/or electrical remodeling.

METHODS

Propofol-anesthetized dogs were subjected to serial magnetic resonance imaging (MRI) measurements, which were preceded by determination of the electrophysiological parameters with the animals being conscious (see below).

Animal handling was in accordance with the Dutch Law on Animal Experimentation and the European Directive for the Protection of Vertebrate Animals Used for Experimental and Other Scientific Purposes (86.609/EU). The experiments were approved by the Committee for Experiments on Animals of Maastricht University and by the Committee for Infections of Academic Hospital Maastricht.

Preparation of the dogs. Six adult mongrel dogs (22–30 kg) of either sex were selected for the experiments after they had been tested in and accommodated to the laboratory environment. After two control MRI examinations in sinus rhythm (SR), CAVB was induced through a right-sided thoracotomy.

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under complete general anesthesia using an injection of 37% formaldehyde at the AV junction. For details of this procedure, we refer to an earlier study (34). During this operation, an endocardial RV electrode (Vitatron, Slimtine ISP13; Dieren, The Netherlands) was inserted and exteriorized through the back of the neck to pace the dog. This specific lead was chosen after preliminary tests, which did not show interference with the MR environment. Proper care was taken before and after the experiments.

MRI technique. The anesthesia regimen used for the MRI experiments consisted of acepromacine (0.04 mg/kg) and was followed after 30 min by an intravenous bolus of propofol (3 mg/kg). A continuous infusion of propofol (0.3 mg·kg⁻¹·min⁻¹) was continued thereafter. No artificial ventilation had to be applied.

MR imaging was performed using a 0.5-T Gyroscan NT whole body scanner (Philips; Best, The Netherlands). With the use of a survey consisting of 3 × 9 images in three orthogonal planes, the heart position was identified. Subsequently, a stack of 9-mm-thick contiguous slices, with a field view of 300 mm, was acquired, covering both ventricles in short-axis views. A cardiac-triggered gradient echo multi-slice-multiphase (CINE) sequence with a repetition time (TR) of 1,200 ms in CAVB dogs (equal to a paced heart rate of 50 beats/min) and a TR of ~600 ms in SR dogs, with an echo time (TE) of 9.4 ms and a 40° flip angle, was used. Each slice was imaged in 20 phases during the cardiac cycle. The resulting images were evaluated using the MR Analytical Software System (MASS) program (version 3.0, Laboratory for Clinical and Experimental Image Processing, Department of Diagnostic Radiology, LUMC, Leiden, The Netherlands).

The lower detection limit of the MR trigger system used was 40 beats/min; therefore, an external pacemaker (5880A, Medtronic; Maastricht, The Netherlands) set at 50 beats/min was applied in all cases after CAVB. Fixed-rate pacing had the additional advantage of the same heart rate in all MRI images obtained after CAVB. On one occasion, the heart was triggered at a very high rate by the MR gradient system, needing relocation of the external connecting cable in the scanner. No other adverse effects were noted in connection with pacing in the MR environment. The duration of the MR examinations was in the order of 30–40 min, including positioning of the animal.

Electrophysiological measurements. Directly after the AV block procedure, the dog was paced at a cycle length (CL) corresponding to the previous SR to overcome the negative effects of anesthesia and thoracotomy. After 1 day, this paced rhythm was stopped, and the first electrophysiological measurements after CAVB were performed with the conscious dog lying quietly on the floor. Six surface electrocardiogram (ECG) leads were simultaneously registered and stored digitally using a sample frequency of 1 kHz. In this series, we chose to measure Q-T time at not only the idioventricular rhythm (IVR) but also at a paced rhythm of 1,000 ms (60 beats/min) to keep the activation fronts and QRS morphology constant.

Time intervals. The autopsy findings in dogs with CAVB from earlier studies in our laboratory showed a similar heart weight-to-body weight ratio after 2 wk of CAVB up to 29 wk of CAVB (Fig. 1). Therefore, the first 3 wk of CAVB were chosen as the primary time of interest, with measurements at SR (twice) and 6, 14, and 21 days after creation of CAVB. In most dogs, additional measurements were made: in three dogs at 2 and 10 days and in two dogs at 5 and 7 wk after creation of CAVB. The electrophysiological measurements were performed for up to 7 wk.

Data acquisition and analysis. After the MRI images were transferred to a UNIX workstation (SUN, Microsysten, data were analyzed with the MASS program from base to apex. With this program, the endocardial and epicardial contours of the heart were manually traced with an optical mouse (Fig. 2). In the LV, automatic contour detection was available, which still could be corrected manually (29a). The papillary muscles were included in the endocardial contour, because they contribute to the ventricular mass. Epicardial fat was carefully excluded. The LV was measured until the appearance of the aorta, whereas the RV was determined until the start of the atria. Mass was calculated as equal to the epicardial volume minus the endocardial volume × 1.05.

An additional feature of the program allows measurement of LV wall thickness (WT) using a modified centerline method. The first (manually drawn) line was put at the posterior side of the septum, going to the left posterior wall. With this program, the endocardial and epicardial surfaces were divided in three parts: LV posterior wall, LV anterior wall, and septum. The width of the RV free wall was determined at end diastole. To evaluate the accuracy of this method, a similar...
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Technique was also applied for the LV free wall and compared with the automatic WT calculation. With the use of the endocardial volume, the end-diastolic (EDV) and end-systolic volumes (ESV) were measured. Using these calculations, the ejection fraction (EF) and cardiac output [equal to (EDV – ESV) × heart rate] could be determined.

With the use of a custom-made computer program (ECG View, University of Maastricht, Maastricht, The Netherlands) with an adjustable time scale and gain, the following parameters were measured offline: 1) CL of the IVR, 2) Q-T time (lead II) during IVR and at 1,000 ms pacing, and 3) P-P intervals. The data reported are the means of at least 5 consecutive beats (Q-T and CL of IVR) and at least 10 beats for the P-P interval.

Autopsy. After the series were completed, all dogs were euthanized, and the hearts were stored in formalin. After at least 1 wk of storage, the atria were removed, and the ventricular weight was determined by dividing the heart into the RV and LV (including septum) using slices of ~1 cm starting at the base of the AV ring. Furthermore, autopsy data of a group of 84 CAVB dogs and 31 SR dogs were used to serve as an external reference (Fig. 1). From that reference group, nine dogs (4 CAVB and 5 SR dogs) were selected in which LV and RV tissue were immediately frozen in liquid nitrogen and stored at ~80°C until mRNA analysis.

Analysis of the angiotensin type 1 receptor mRNA. Earlier, we (34) reported that the angiotensin II (ANG II) plasma level was temporarly elevated during CAVB with its peak around 2 wk and normalization toward control at 6 wk (34). ANG II is the determinant factor of the renin-angiotensin system (RAS). Most of its effects (including hypertrophy) are mediated by the ANG II type 1 (AT1) receptor (for a review, see Ref. 13). Therefore, we assessed the AT1 receptor mRNA expression in LV and RV tissue using a Light Cycler (Roche).

This novel technique enables semiquantitative analysis of low-abundance mRNA using real-time PCR.

Total RNA was isolated from the tissue using a CsCl2 gradient and ultracentrifugation. Genomic DNA contamination of the samples was largely eliminated by digestion with DNase I (Promega). cDNA was synthesized using an optimized reverse transcription protocol with an oligodT-VN anchored primer (Lekanne Deprez, Academic Medical Centre Amsterdam, Amsterdam, The Netherlands, personal communication). Real-time PCR was performed with a FastStart DNA Master Sybr Green 1 kit (Roche) and dog-specific AT1 receptor primers (17).

As an internal standard, dog-specific glyceraldehyde-3-phosphate dehydrogenase primers were used to enable correction for total RNA amounts. For all comparisons (SR LV vs. CAVB LV, SR RV vs. CAVB RV, and LV vs. RV), a separate run was performed.

Statistics. Pooled data are expressed as means ± SD unless otherwise indicated. Analysis of variance, followed by Student’s t-tests, was applied to compare the data after CAVB in respect to SR values. The AT1 receptor differences were tested by Student’s t-tests. Correlation analysis was performed using Primer of Biostatistics software. Differences were considered significant if \( P < 0.05 \).

RESULTS

Validation of the MRI measurements. The two mass measurements performed during SR were comparable and, therefore, taken together as one control; the maximum difference between measurements was 7.4 ± 5.2 g for the LV and 4.5 ± 3.7 g for the RV. These values were far below the expected increase in mass of ±20–25 g in both ventricles (34), confirming other studies (6, 20) that showed that a reliable prediction of the increase in mass was possible. The autopsy data and the MRI results showed also a good correlation (\( r = 0.98, P < 0.001 \)).

Development of hypertrophy in time. CAVB caused a gradual increase in the mass of both ventricles (Table 1 and Fig. 3, top left). At 6 days (LV) and 14 days (RV), respectively, this increase in ventricular weight became significant. After 14–21 days, the progression seemed to stabilize; no further increase in mass was found, which was confirmed in the two dogs with a longer follow-up. A representative example of the long-term alteration in LV mass is shown in Fig. 4 (top) in which the extended MRI measurements were used.

With the use of MRI to calculate the increase in the ventricle-to-body weight ratio, these dogs showed an increase in the LV-to-body weight ratio from 4.5 ± 1 to 6.0 ± 0.5 kg/kg, whereas the RV-to-body weight ratio increased from 1.5 ± 0.1 to 2.1 ± 0.1 kg/kg \( (P < 0.05) \). Similar results were obtained in the much larger autopsy groups; during SR, the LV-to-body weight ratio was 4.3 ± 0.8 kg/kg, and the RV-to-body weight ratio was 1.6 ± 0.3 kg/kg. After CAVB, the LV-to-body weight ratio increased to 5.8 ± 1 kg/kg, and the RV-to-body weight ratio increased to 2.6 ± 0.6 kg/kg \( (P < 0.001) \) for both.
Nature of hypertrophy (WT and volumes). WT measurements at SR showed very comparable results, e.g., the consecutive measurements of the RV end-diastolic WT differed by 0.1 mm. With the use of the automatic detection method (Fig. 2), there was no increase in LV end-diastolic WT after CAVB (Table 1 and Fig. 3, top right). Manual measurement of LV end-diastolic WT showed the same quantitative results: 10.5 ± 1.1 mm at SR vs. 10.2 ± 1.1 mm after 3 wk of CAVB (not significant). This in contrast to the LV end-systolic WT, which increased considerably (P < 0.01; Table 1) and homogeneously; both absolutely and relatively, there was no difference in the WT increase in the different parts of the LV (Table 1).

The RV end-diastolic WT showed a significant increase from 5.8 ± 0.6 to 7.9 ± 0.9 mm at 21 days of CAVB (Table 1 and Fig. 3, top right).

The assessment of the EDV showed a clear increase in LV EDV (Table 2) and RV EDV (from 58 ± 25 to 73 ± 12 ml, P < 0.05).

Comparison of RV and LV hypertrophy. The ventricles responded differently to volume overload; the relative increase in mass was much greater for the RV, most likely due to the important increase in WT (Fig. 3, top right and top left). The LV increased in mass because of a longitudinal increase in cavity size, as assessed by the number of slices covering the LV (Table 1), indicating an increase from apex to base of maximal 9 mm compared with SR. In Fig. 5, a transversal view of the heart at SR and 21 days of CAVB shows this differential structural adaptation of the ventricles.

Molecular findings. In the group of SR dogs, there was similar AT1 receptor mRNA expression in the RV and LV (Fig. 6). After CAVB, the AT1 receptor mRNA expression of the RV was significantly larger than that in the RV of SR dogs, whereas the LV did not show a change (Fig. 6).

Functional parameters. The structural adaptations were accompanied by an increase in LV EF from 68 ± 6 to 86 ± 5% at 14 days of CAVB (Table 2). In this way,

Table 1. Changes in ventricular dimensions and weight as obtained by MRI analysis

<table>
<thead>
<tr>
<th></th>
<th>SR</th>
<th>CAVB Day 6</th>
<th>CAVB Day 14</th>
<th>CAVB Day 21</th>
<th>% Increase</th>
</tr>
</thead>
<tbody>
<tr>
<td>LV mass, g</td>
<td>116 ± 11</td>
<td>136 ± 16*</td>
<td>143 ± 12*</td>
<td>147 ± 9*</td>
<td>27</td>
</tr>
<tr>
<td>RV mass, g</td>
<td>40 ± 2.8</td>
<td>49 ± 10.2</td>
<td>55 ± 5.6*</td>
<td>56 ± 3.9*</td>
<td>40</td>
</tr>
<tr>
<td>LV end-systolic WT, mm</td>
<td>17.2 ± 1.9</td>
<td>20.7 ± 2.1*</td>
<td>23.6 ± 3.2*</td>
<td>22.5 ± 3.9*</td>
<td>30</td>
</tr>
<tr>
<td>Posterior</td>
<td>18.3 ± 2.2</td>
<td>23.7 ± 3.2</td>
<td>24.7 ± 3.5</td>
<td>23.1 ± 3.1</td>
<td></td>
</tr>
<tr>
<td>Septum</td>
<td>16.9 ± 1.4</td>
<td>20.0 ± 1.8</td>
<td>24.2 ± 4.1</td>
<td>22.9 ± 5.3</td>
<td></td>
</tr>
<tr>
<td>Anterior</td>
<td>16.5 ± 1.7</td>
<td>17.5 ± 1.1</td>
<td>21.7 ± 2.7</td>
<td>21.2 ± 3.5</td>
<td></td>
</tr>
<tr>
<td>LV end-diastolic WT, mm</td>
<td>11.6 ± 1.7</td>
<td>11.7 ± 1.5</td>
<td>12.4 ± 0.8</td>
<td>12.2 ± 1.4</td>
<td></td>
</tr>
<tr>
<td>LV longitudinal axis, no. of slices</td>
<td>6.5 ± 0.4</td>
<td>7.4 ± 0.4*</td>
<td>7.2 ± 0.2*</td>
<td>7.4 ± 0.4*</td>
<td>13</td>
</tr>
<tr>
<td>RV end-diastolic WT, mm</td>
<td>5.8 ± 0.6</td>
<td>7.7 ± 1.8</td>
<td>8.3 ± 0.6*</td>
<td>7.9 ± 0.9*</td>
<td>34</td>
</tr>
<tr>
<td>Posterior</td>
<td>18.3 ± 2.2</td>
<td>23.7 ± 3.2</td>
<td>24.7 ± 3.5</td>
<td>23.1 ± 3.1</td>
<td></td>
</tr>
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<td>7.2 ± 0.2*</td>
<td>7.4 ± 0.4*</td>
<td>13</td>
</tr>
<tr>
<td>RV end-diastolic WT, mm</td>
<td>5.8 ± 0.6</td>
<td>7.7 ± 1.8</td>
<td>8.3 ± 0.6*</td>
<td>7.9 ± 0.9*</td>
<td>34</td>
</tr>
</tbody>
</table>

Values are means ± SD. MRI, magnetic resonance imaging; CAVB, complete atrioventricular (AV) block; SR, sinus rhythm; LV and RV, left and right ventricle, respectively; WT, wall thickness. * P < 0.05 vs. SR.

Fig. 3. Temporal behavior of structural, electrical, and mechanical remodeling. Top: time course of the structural changes. Top left: relative increase in LV and RV mass. Moreover, it can be seen that the structural remodeling of the RV is greater than that in the LV. Top right: temporal behavior of the relative increase in LV and RV end-diastolic WT (EDWT), with no change in LV EDWT and a clear increase in RV EDWT. Bottom left: relative increase in Q-T time at 1,000 ms pacing. Bottom right: temporal changes in cardiac output (CO). The electrical remodeling parallels the structural adaptation in time, whereas CO returns to pre-CAVB circumstances.
the cardiac output of the CAVB dogs was maintained after an initial reduction (Table 2 and Fig. 3, bottom right). After 2 wk of CAVB, the LV mass-to-EDV ratio was similar to the ratio at SR (Table 2). The RV mass-to-EDV ratio at SR (0.77 g/ml) was also comparable with that at 3 wk of CAVB (0.79 g/ml).

Electrophysiological measurements. Under anesthesia, the Q-T interval in SR dogs (CL of 505 ± 70 ms) amounted to 250 ± 20 ms. The CL of IVR and the P-P interval of the conscious dogs were stable over the week after creation of AV block (Table 3). The Q-T at IVR increased significantly from 260 ± 22 ms at day 1 to 285 ± 28 ms at day 21. With the paced rhythm (1,000 ms), an increase in Q-T time and hypertrophy reached their maximum at 2 wk. There is a clear increase in Q-T time during the first 2 wk, which reached a plateau at ∼14–21 days. Both the paced Q-T time and hypertrophy reached their maximum at 2 wk.

After 14–21 days, the Q-T interval increased significantly from 285 ± 28 ms at day 21 to 335 ± 35 ms (P < 0.01) (Fig. 4, bottom left). After 14–21 days, the Q-T interval stabilized, as illustrated in Fig. 4 (bottom left and bottom right). No changes were observed in QRS width during pacing.

**DISCUSSION**

In this study, a serial analysis of the LV and RV adaptation processes after bradycardia-induced volume overload revealed that structural remodeling starts very quickly and is completed within 2–3 wk after CAVB. In parallel, i.e., in the same time frame, electrophysiological (increase in Q-T time) and mechanical changes (maintaining cardiac output) occurred. The degree of the hypertrophy differed between the ventricles.

**Dog model of AV block.** Comparing SR/acute AV block with CAVB (>4 wk), major adaptations have been described at the structural, functional, and electrophysiological levels, which all have been confirmed in isolated myocytes obtained from both ventricles (24, 32). In the beginning of this century, it had already been noted that CAVB in dogs leads to biventricular hypertrophy (7), which is not dependent on the duration of CAVB (Fig. 1) (3, 25, 29, 36) and develops without an increased collagen fraction (fibrosis) and with a normal capillary-to-myocyte ratio (34).

In acute AV block, cardiac output falls to ∼65–75% of the original SR value (5, 25). This reduction is proportional with the severity of the ensuing bradycardia (3, 26). Less agreement exists in the literature in regard to the mechanical performance during CAVB; the first studies (3, 25) described the CAVB dog as a model for heart failure, whereas later studies reported compensated cardiac function in the majority of dogs. In our experience, improved contractile function at the IVR has been observed after 6 wk of AV block (5).

In many laboratories, the AV block dog is used to study ventricular arrhythmias. The response of the dogs to interventions (e.g., drugs) is dependent on the duration of AV block, which is reproducible after 2–3 wk of CAVB (35), suggesting that electrical adaptations are completed within this time period. Electrical remodeling consists of an increased interventricular dispersion (34), which has been related to ventricular arrhythmias, more specifically torsade de pointes (31, 33, 34).

**Methodological considerations.** For this study, we were interested in 1) the time course of the structural adaptation process of both ventricles after CAVB, 2) the comparison of this process in time with mechanical and electrophysiological adaptations, and 3) changes at the executor site of the RAS system, that being the AT1 receptor. Data from the RV are difficult to obtain echocardiographically and with the MRI technique are limited to SR dogs (20). To our knowledge, this is the first study describing the structural adaptations of the RV over time.

**Table 2. Hemodynamic values as obtained by MRI analysis of the LV**

<table>
<thead>
<tr>
<th>Day 0 (SR)</th>
<th>Day 6</th>
<th>Day 14</th>
<th>Day 21</th>
</tr>
</thead>
<tbody>
<tr>
<td>EDV, ml</td>
<td>63 ± 14</td>
<td>87 ± 12</td>
<td>87 ± 14*</td>
</tr>
<tr>
<td>EF, %</td>
<td>68 ± 6</td>
<td>77 ± 9</td>
<td>86 ± 5*</td>
</tr>
<tr>
<td>CO, l/min</td>
<td>4.0 ± 0.7</td>
<td>3.5 ± 0.2</td>
<td>3.8 ± 0.3</td>
</tr>
<tr>
<td>Mass/EDV</td>
<td>1.8 ± 0.4</td>
<td>1.6 ± 0.3</td>
<td>1.7 ± 0.3</td>
</tr>
</tbody>
</table>

Values are means ± SD. EDV, end-diastolic volume; EF, ejection fraction; CO, cardiac output. *P < 0.05 vs. day 0 (SR).
Time course of structural and functional changes in the CAVB dog. Including the additional time points at 2 and 10 days after CAVB, we were able to demonstrate the quickness by which biventricular hypertrophy evolves. The clear increase in LV mass after 2 days of CAVB (Fig. 4) is in contrast to other data (25, 36) in which the hypertrophy became evident after 10–18 days and clearly manifested between 3 and 7 mo. Whether the RV behaves in a similar time fashion is less clear. Imaging the RV in dogs still is complex due to its position in the thorax. This is most probably the reason that the increase in RV mass as measured with MRI after CAVB was somewhat less compared with that as assessed in autopsied animals (34). Still (Figs. 1, 3, and 4), this remodeling process of both ventricles seems to stabilize after 14–21 days. At least, the majority in growth (90% completion) is seen within 14 days.

In both ventricles, the cardiac mass-to-EDV ratio at chronic CAVB is similar to that in SR, implicating eccentric hypertrophy. This ratio (often used as an indicator for ventricular dilatation) decreases from 1.6 to 1.13 g/ml in dogs with heart failure (6, 14).

Other canine models of volume overload and hypertrophy. Other models of LV overload, such as mitral valve regurgitation (MVR), have reported lower or similar structural adaptations: LV mass increases to 5.0–5.6 g/kg (19, 22, 37, 38). In MVR, the time course is much slower; after 3 wk, only half of the total growth (±10 g) has taken place, and it takes ~9 wk before a steady state in LV mass is reached (19). Right-sided changes in mass are absent (37) or not addressed in these articles.

Solitary RV volume overload is not often used in dogs. In one model, a connection was made between the right atrium and the pulmonary artery (16). The RV WT increased by 35% after 30 days and the RV-to-body weight ratio increased to ±2.1 g/kg (16), which is somewhat lower than the observed changes in CAVB.

Creation of an arteriovenous shunt is an example of volume overload that results in biventricular hypertrophy. The LV mass reaches similar or higher values as that in CAVB, ranging from 5.8 to 6.6 g/kg (18, 22), whereas the RV increased to ±2.2 g/kg (18, 22). Similar to our observations, the growth in mass is mainly due to an increase in length of the LV cavity (22) without an increased LV end-diastolic WT. This increase in base-to-apex length was first documented after 2–4 wk (22), suggesting a slower adaptation rate. To our knowledge, information about RV end-diastolic WT in AV shunt has not been reported.

Both MVR and AV shunt canine models lack data to make a full temporal comparison of development of hypertrophy. Still, it seems plausible to say that vol-

Table 3. Electrophysiological parameters after AV block during IVR

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Day 1</th>
<th>Day 6</th>
<th>Day 14</th>
<th>Day 21</th>
</tr>
</thead>
<tbody>
<tr>
<td>IVR, ms</td>
<td>1,260 ± 230</td>
<td>1,220 ± 225</td>
<td>1,105 ± 120</td>
<td>1,160 ± 145</td>
</tr>
<tr>
<td>PP, ms</td>
<td>370 ± 90</td>
<td>370 ± 90</td>
<td>360 ± 55</td>
<td>370 ± 60</td>
</tr>
<tr>
<td>Q-T time, ms</td>
<td>260 ± 22</td>
<td>275 ± 26*</td>
<td>280 ± 30*</td>
<td>285 ± 29*</td>
</tr>
</tbody>
</table>

Values are means ± SD. IVR, idioventricular rhythm; PP, P-P interval. *P < 0.05 vs. day 1.
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Ufumble overload alone does not explain the rapid time frame of growth in CAVB. When we speculate about other possible causes, we infer that 1) volume overload in CAVB may result in higher (differential) ventricular wall stresses and by that in higher neurohumoral responses, or 2) the prolonged diastolic interval creates more time for growth. Part of the electrical adaptation in MVR and AV shunt is an increase in heart rate.

Background of hypertrophy. The hypertrophy is initiated by mechanical factors and thereafter amplified by neurohumoral factors, such as adrenergic stimuli, RAS, endothelin, and insulin-like growth factor.

In the present study, we restricted ourselves to the effect of CAVB on AT1 receptor mRNA expression, because ANG II is the acting factor in the RAS and so far all its major effects are regulated via the AT1 receptor (13). A previous study (34) from our group showed a temporarily increase in ANG II plasma levels after CAVB creation, which were normalized at 6 wk of CAVB. In the canine heart, ANG II is derived from different sources: plasma or local synthesis by either angiotensin-converting enzyme or chymase from either locally or plasma-derived angiotensinogen (6, 17, 35).

In agreement with Lee et al. (17), SR dogs had a similar ventricular AT1 receptor mRNA expression. In dogs with CAVB, the RV AT1 receptor mRNA increased compared with SR, which could be an explanation for the observed difference in degree of hypertrophy of the RV. Moreover, the increase in AT1 receptor shows an important role for the RAS system in the development of the hypertrophy after CAVB, although other factors cannot be ruled out.

An equal or upregulated AT1 receptor mRNA expression is in contrast to dogs with volume overload due to MVR (6, 27) and dogs with right-sided hypertrophy after pulmonary banding and tricuspidal insufficiency (17), in which the AT1 receptor mRNA content decreased. This difference in AT1 receptor response could be due to differences in models and/or compensated hypertrophy in CAVB versus heart failure.

Also, in patients with heart failure, the density of AT1 receptors was decreased (1, 11), and the lowest values were found in tissue of patients with the worst function (1). The noninfarcted region of the heart in these patients showed an increase in the AT1 receptor (21). When speculating, we hypothesize that AT1 receptors are upregulated in compensated hypertrophy and reduced in heart failure.

Long-term electrophysiological adaptations We observed a temporal increase in Q-T time during IVR, which reached a maximum ~2–3 wk after AV block (Table 3). Similar results were obtained when the CL and activation sequence were controlled by pacing from the RV endocardial electrode (Fig. 4), indicating that this increase in repolarization time represents true electrical remodeling. The Q-T time increased more during the paced rhythm (±10% IVR vs. ±20% at 1,000 ms), which is most probably related to sequential activation of the RV and LV. The paced Q-T increase of 18% is well in line with serial measurements performed during acute and chronic AV block in anesthetized dogs (34). Q-T time is a global reflection of repolarization, which in anesthetized dogs shows the best correlation with LV APD having an equation close to unity (31). Therefore, the increase in Q-T time is most probably due to the changes in repolarization of the LV. Both in vivo and at a cellular level, we (32, 34) have demonstrated that the RV APD increases less than the LV APD in CAVB dogs (increased dispersion). This is despite the fact that the RV hypertrophy is more pronounced, suggesting that hypertrophy is not the sole contributor to electrical remodeling.

Hypertrophy is linked with a prolonged repolarization and an increased incidence of ventricular arrhythmias and sudden cardiac death in patients (9, 10, 12, 39). These risks are not restricted to (severe) pathological circumstances in which heart failure is present but also occur in physiological states, such as in the heart of endurance athletes with compensated hypertrophy (23) or as described in this animal model with improved inotropic performance (maximal LV rise in pressure over time) at slow rates (5).

Investigations combining information about structural, functional, and electrical measurements are relatively scarce and often restricted to observations related to the increased sensitivity to (pacing induced) arrhythmias after interventions, such as ischemia or drugs (2, 15). Moreover, exact information concerning the sequence and nature of the time-related electrophysiological process in hypertrophy are lacking. In this study, we showed that the increase in Q-T time parallels the process of hypertrophy and mechanical adaptation. Whether they are caused by similar signaling pathway or different pathways has yet to be elucidated using prevention or regression studies, in which AT1 receptor blockade should be the first to be tested.

Technical limitations. With regard to MR measurements, the determination of the RV size and WT is not easy. Still, based on the similar outcome of the repetitive measurements in SR and the correlation with the autopsy data, we are confident that the MRI accurately described the time process of growth. The quantitative difference between the automatic and the manual WT measurements is due to the inclusion of papillary muscles in the former method.

The possible role of the shifting activation patterns of the IVR during the day has not been assessed. Changing the ventricular activation pattern by LV free wall pacing has been reported to result in an increase in LV mass at the region distant from the electrode, which took ~3 mo before completion. In contrast, at the pacing site wall thinning was found (30). In our data, the LV wall thickness at end systole increased homogeneously, indicating that activation did not influence the process of hypertrophy.

In conclusion, electrical, mechanical, and structural adaptation processes after a sudden bradycardia-induced volume overload are rapid and parallel. Within 3 wk, all studied processes have reached a plateau. The degree of hypertrophy was greater in the RV, which was associated with an increase in AT1 receptor mRNA expression in this ventricle after CAVB.
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